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PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants:	Zhao, et al.		
Serial No.:	09/469,485	Case No.: 20369Y	Art Unit: 1648
Filed:	December 22, 1999		Examiner: Foley, Shanon A.
For:	IMPROVED RECOMBINANT HEPATITIS B SURFACE ANTIGEN		

The Honorable Commissioner of Patents and Trademarks
Washington, D.C. 20231
Attention: Board of Patent Appeals And Interferences

APPELLANT'S REPLY BRIEF UNDER 37 CFR 1.193

Sir:

Appellant requests consideration of enclosed Reply Brief in response to the Examiner's Answer mailed June 16, 2004, Confirmation No. 5022. The Reply Brief is being filed in triplicate.

If any time extensions are needed for the timely filing of the Appellant's Reply Brief, Appellant petitions for such extensions and authorizes the charging of Deposit Account 13-2755 for the appropriate fees.

Respectfully submitted,

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I hereby certify that this correspondence is being deposited with the United States Postal Service as first class mail in an envelope addressed to: Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313-1450, on the date appearing below.

MERCK & CO., INC.
By Kathy Krajcik Date 8/16/04

Date: 16 August 2004

BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES

Application Number: 09/469,485

Filing Date: December 22, 1999

Appellant(s): Zhao, et al.

Examiner: Foley, Shanon A.

APPELLANT'S REPLY BRIEF

Response to Examiner's Answer

Appellant's response to the Examiner's Answer is provided below. The section numbering corresponds to the issues presented in Appellant's Appeal Brief and Examiner's Answer.

(1) REAL PARTY IN INTEREST

a. The real party in interest in this appeal is Merck & Co., Inc. ("Merck"), as described in the Appeal Brief dated April 5, 2004.

(2) RELATED APPEALS AND INTERFERENCES

a. In the Examiner's Answer filed 6/16/2004, the Examiner states the following:

b. The brief does not contain a statement identifying the related appeals and interferences which will directly affect or be directly affected by or have a bearing on the decision in the pending appeal is contained in the brief. Therefore, it is presumed that there are none. The Board, however, may exercise its discretion to require an explicit statement as to the existence of any related appeals and interferences.

c. There are presently no related appeals or interferences.

(3) STATUS OF CLAIMS

a. Claims 1-20 are currently pending in the application at the time of the filing of the Notice of Appeal, with Claims 1-7 withdrawn from consideration. This appeal involves Claims 8-20.

(4) STATUS OF AMENDMENTS

a. The amendments submitted August 7, 2003 have been entered and appear in the claims on appeal. There are no outstanding amendments, as noted in the Appeal Brief dated April 5, 2004.

(5) SUMMARY OF INVENTION

a. In the Examiner's Answer filed 6/16/2004, the Examiner states the following:

b. The summary of invention contained in the in the brief is deficient because the summary encompasses limitations that are not present in the claims on appeal.

The invention claimed is drawn to increasing the antigenicity of a recombinant hepatitis B surface antigen (rHBsAg) by filtering rHBsAg from a cell culture, adding a redox buffer, adjusting the temperature to 34° to 38° C and incubating the recombinant rHBsAg at the adjusted temperature for 40 to 240 hours.

c. Applicant redraws the Summary of the Invention as follows:

The present invention provides a method of making an improved recombinant hepatitis B surface antigen, rHBsAg, that has a higher specific antigenicity than previously known rHBsAg.

(6) STATEMENT OF ISSUES

Examiner acknowledges that Appellant's statement of the issues in the brief is substantially correct. Changes in Section II: The brief states that claim 19 is "dependent on Claim 8 through Claim 18". However, claim 19 is dependent on claim 18.

Appellant's response to Examiner's corrections in Issue II is as follows:

II. Whether Claim 17, dependent on Claim 8 and further comprising the steps of adding aluminum adjuvant and co-precipitating; Claim 19, dependent on Claim 18, which Claim 18 is dependent on Claim 8, limited to a range of incubation times and further comprising the steps of adding formalin and incubating, and the steps of adding aluminum adjuvant and co-precipitating; and Claim 20, dependent on Claim 8 but limited to certain incubation times and further comprising the steps of adding aluminum adjuvant and co-precipitating, are unpatentable under 35 U.S.C. § 103 over Builder *et al.* and Valenzuela *et al.*, as applied to Claims 8-16 in view of Petre *et al.*, WO 93/24148 A1.

(7) GROUPING OF CLAIMS

Appellant's brief includes a statement that claims 8-20 do not stand or fall together and provides reasons as set forth in 37 CFR 1.192(c)(7) and (c)(8).

(8) CLAIMS APPEALED

Claims 8-20 are on Appeal. In view of the errors cited by the Examiner, and the Examiner's incorrect statementof Claim 8, a new Appendix of the Claims is attached hereto.

ARGUMENT

I. The Examiner does not properly apply the Builder reference in stating that Claims 8-16 are patentable under 35 U.S.C. § 103 over Builder *et al.*, US 4,620,948 in view of Valenzuela *et al.*, 1979. Nature 280:815-819.

In paragraph (10) of the Examiner's Answer the Examiner states the above cited rejection. Appellant has responded to this rejection in the Appeal Brief. However, in reply to the Examiner's Answer, Appellant submits the following additional argument.

Claims 8-16 recite a method of making rHBsAg starting from soluble sterile filtered rHBsAg. Applicant summarizes the Examiner's position to be that Valenzuela provides motivation for one of skill in the art to make properly folded HBsAg and that Builder provides a method of refolding proteins into an active conformation.

In the paragraph bridging pages 4 and 5 of the Examiner's Answer the Examiner again makes clear his basic miscomprehension of the Builder reference. The Examiner states

"The reference exemplifies ways to recover biologically active protein from cell culture by adding a weak denaturing solution. Builder et al teach adding 10mM GSH: 1mM GSSG to solubilized protein and incubating the mixture..." (underlining be Examiner)

The Examiner's underlining illustrates the Examiner's miscomprehension of the Builder reference. The reference teaches adding the denaturing solution to insoluble protein to refold it. Appellant reviewed this issue in the Appeal Brief under Issue I and incorporates those arguments here.

In support of the Examiner's argument, the Examiner cites specific teachings of the Builder reference. However, each and every citation is irrelevant to the presently claimed methods starting with a soluble protein as recited in Claim 8. Claim 8 requires the provision of a soluble protein as a starting material. That limitation, added by Amendment on October 10, 2002, in itself should remove Builder's method of refolding insoluble protein as a reference. Nonetheless, the Examiner persists in citing the inapplicable methods of Builder.

In particular, the Examiner cites Cols 4-5, a treatment for a refractile, insoluble protein. Similarly, the citations to Col 16, lines 28-55 and Col 17, lines 24-56 are methods to

follow on after the complete breaking of all sulfur bonds in a sulfitolysis reaction (Col 16, lines 7-27, Col 17, lines 16-31). The Examiner's citation to Col 20, lines 28-57 is to some purification steps that can be used before denaturation and renaturation of insoluble protein. Builder teaches these methods as part of a procedure for handling "heterologous precipitated (refractile) protein" (Col 19, lines41-42) which have been recovered from the insoluble pellet resulting when "the insoluble material is separated from soluble proteins preferably by centrifugation and the supernatant is removed. The supernatant contains primarily the host proteins and is discarded" (Col 15, lines 50-53, underlining added).

Clearly, the Examiner is improperly citing the Builder reference by applying the reference in direct opposition to it's own teachings. Builder teaches that the methods are for insoluble proteins. The present claims require soluble protein as the starting material.

Clearly, as recited in the Appeal Brief, Builder, *et al.*, teaches a method designed, in Builder's own words quoted below, for refolding insoluble proteins which have been produced by host cells as refractile bodies. To wit, Builder *et al.*, states:

"A large number of human, mammalian, and other proteins, including, for example, human growth hormone, (hGH) bovine growth hormone (bGH) and a number of interferons have been produced in host cells by transfecting such cells with DNA encoding these proteins and growing resulting cells under conditions favorable to the expression of the new heterologous protein. Viral coat proteins, such as capsid proteins of foot and mouth disease (FMD) virus and the surface antigenic protein of hepatitis B virus (HBsAg) are still other examples of heterologous proteins which have also been produced in suitable recombinant DNA engineered hosts. The **heterologous protein is frequently precipitated** inside the cell, and constitutes a significant portion of the total cell protein." (lines 18-32, **emphasis** added).

Further, Builder *et al.*, teach:

"Various heterologous proteins expressed in bacterial host cells, for example, pGH, hGH, and viral coat proteins such as a fusion protein with FMD virus, protein and HBsAg form refractile bodies **to a greater or lesser extent** under commonly found culture

conditions. Certain other proteins such as immune interferon (IIF) and leukocyte interferon (LeIF) are more soluble in the cytoplasm. (Fibroblast interferon (FIF) is, however, refractile in host culture.)" (col. 6, lines 48-56, **emphasis** added)

Moreover, Builder *et al.*, states:

"The invention herein is directed to procedures which are useful in isolating, purifying, and, if necessary, reactivating proteins which appear in host cells in the form of "refractile bodies". Part of the invention concerns methods which encourage such refractile body formation; **however, the procedures for protein recovery and activation disclosed herein are intended to be specifically applicable to such refractile proteins.**" (col 6, lines 30-37, **emphasis** added)

Therefore, the only manner in which the Examiner can state that Builder is relevant is to ignore the direct and explicit teaching of the reference, as cited by the Examiner and as quoted above, that the method is designed for use with insoluble and refractile proteins.

The starting material required by Claim 8 is soluble. However, the Examiner seems to have missed this fact as stated at page 11, lines 5-6

"However, how the starting material is ultimately provided is not a recited element in the claims." (emphasis original by the Examiner).

Further, at page 5, lines 13-15, the Examiner states that one would have been motivated to combine Valenzuela with Builder. However, the Examiner again fails to recognize that Valenzuela does not produce an insoluble protein in need of refolding. Therefore, one would not be motivated to apply the teachings of Builder where that teaching does not apply.

Appellant sees the Examiner as continuing to present an improper rejection in which the Builder reference is cited in direct opposition to it's clear teaching, and in which the Examiner has miscomprehended the importance, and even the existence, of the limitation in Claim 8 that the starting material be soluble. Therefore, Appellant respectfully requests that the Examiner's argument be rejected by the Board and that the patentability of the present claims over the prior art of record be confirmed.

II. Claims 17, 19 and 20 are patentable under 35 U.S.C. § 103 over Builder *et al.*, US 4,620,948 and Valenzuela *et al.*, 1979. Nature 280:815-819 in view of Petre et al., WO 93/24148.

Claims 17, 19 and 20 were rejected over Builder *et al.* and Valenzuela *et al.*, in view of Petre *et al.*, WO 93/24148 A1. Applicant incorporates herein all of the comments above as applied to Builder et al., and Valenzuela et al. Because the teaching of adjuvanting cited in Petre et al. does not make up for the deficiency of Builder et al., and Valenzuela et al., as the primary references, the combinations can not provide the basis for a *prima facie* case of obviousness against the patentability of the present claims. Therefore, Applicant respectfully requests that the stated rejection against Claims 17, 19 and 20 be reversed.

III. Claim 18 is patentable under 35 U.S.C. § 103 over Builder et al., US 4,620,948 and Valenzuela et al., 1979. Nature 280:815-819 in view of Even-Chen, US 5,242.812.

Claim 18 was rejected over Builder et al., and Valenzuela et al., in view of Even-Chen, US 5,242,812. Applicant incorporates all of the comments above as applied to Builder et al., and Valenzuela. Because the teaching of the use of formalin cited in Even-Chen does make up for the deficiency of Builder et al., and Valenzuela et al., as the primary references, the combinations can not provide the basis for a *prima facie* case of obviousness against the patentability of the present claims. Therefore, Applicant respectfully requests that the stated rejection against Claim 18 be reversed.

CONCLUSION

Appellant respectfully submits that claims 8-20 are not obvious in view of the cited art. Appellant requests that the Board of Patent Appeals and Interferences reverse the outstanding rejections of claims /under 35 U.S.C. § 103(a).

Please charge deposit account 13-2755 for fees due in connection with this Appeal Brief. If any time extensions are needed for the timely filing of the present Appeal Brief, Appellants petition for such extensions and authorize the charging of deposit account 13-2755 for the appropriate fees. Appellant requests that all rejections be withdrawn and that the invention described be allowed to issue in a patent.

Three copies of this Reply Brief are provided. No fee is due.

Respectfully submitted,

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Date: 16 August 2004

APPENDIX OF CLAIMS ON APPEAL

8. A method of making recombinant hepatitis B surface antigen (rHBsAg) comprising:

a) providing soluble sterile filtered rHBsAg purified from a cell culture,

b) adding a redox buffer to the rHBsAg,

c) adjusting the temperature to 34 to 38 °C,

d) incubating the rHBsAg at 34 to 38 °C for 40 to 240 hours,

wherein the antigenicity of the rHBsAg produced after step d is greater than the antigenicity of the rHBsAg provided in step a.

9. The method of Claim 8 wherein step c is performed before step b.

10. The method according to Claim 8 wherein the redox buffer comprises thiol compounds selected from the group consisting of thiol compounds having a MW less than about 1000 Da and the corresponding disulfide compounds.

11. The method according to Claim 10 wherein the redox buffer is a mixture of at least one thiol compound and at least one disulfide compound.

12. The method according to Claim 11, wherein the ratio of thiol compound to disulfide compound is between about 30:1 and about 1:1.

13. The method according to Claim 12 wherein the concentration of thiol compound is between about 0.05 mM and about 5.00 mM.

14. The method according to Claim 13 wherein the ratio of thiol to disulfide is selected from the group consisting of about 20:1, about 10:1, about 10:4, about 5:1, about 2:1 and about 1:1.

15. The method according to Claim 13 wherein the thiol compound is glutathione and the disulfide compound is oxidized glutathione.

16. The method according to Claim 15 wherein the concentration of glutathione is about 1.0 mM and the concentration of oxidized glutathione is about 0.2 mM.

17. The method according to Claim 8 further comprising the steps of
e) adding an aluminum adjuvant, and
f) co-precipitating the rHBsAg and the adjuvant.

18. The method according to Claim 8 further comprising the steps of
e) adding about 0.01% final concentration of formalin ,
f) incubating the rHBsAg at about 34°C to about 38 °C from about 40 to about 72 hours,
wherein the incubation in step d is from about 40 to about 190 hours.

19. The method according to Claim 18 further comprising the steps of
g) adding an aluminum adjuvant, and
h) co-precipitating the rHBsAg and the adjuvant.

20. The method according to Claim 8 wherein the incubation in step d is about 60 hours and further comprising the steps of
e) adding an aluminum adjuvant, and
f) co-precipitating the rHBsAg and the adjuvant,
wherein step f includes an incubation of about 40 hours.